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Redescription of the alpine *Incurvaria stangei* Rebel, 1903 (Lepidoptera, Incurvariidae), reinstated from Prodoxidae

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http://zoobank.org/282B6590-CADE-4745-AA51-D64DDD4D7303

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Abstract. *Incurvaria stangei* Rebel, 1903 revised combination, placed without justification in Prodoxidae, is redescribed and transferred back to the Incurvariidae. Male and female genitalia of this south-eastern alpine endemic species are illustrated for the first time. DNA barcode sequences as well as morphological traits support the revised family assignment.

Introduction

Incurvariidae and Prodoxidae are two out of formerly five (van Nieukerken et al. 2011), now six (Regier et al. 2015) known families of the superfamily Adeloidea Bruand, 1850. The type-species for Incurvariidae is *Incurvaria masculella* (Denis & Schiffermüller, 1775) and for Prodoxidae *Prodoxus decipiens* Riley, 1880 (Nye and Fletcher 1991). The monophyly of both families is well supported by a morphologically based phylogenetic analysis (Nielsen and Davis 1985) and by recently published molecular studies (Mutanen et al. 2010; Regier et al. 2015).

Incurvaria stangei was described by Rebel (1903) from a single female collected in the Italian Dolomites (Südtirol, Tschierspitze, 31.vii.1902) by G. Stange. Shortly after this discovery a single male was found in the area of Triglav mountain range, currently Slovenia (Rebel 1907). Despite its extraordinary and unmistakable external appearance, this remarkable alpine species of primitive moth remained neglected for more than eight decades. Only in 1988 P. Huemer discovered a specimen and published a re-description, although without examination of genitalic structures (Huemer 1991). Nevertheless, he still treated the species in the family Incurvariidae Spuler, 1898. In contrast, Wojtusiak (1996) in the European Lepidoptera catalogue listed the species under *Lampronia* Stephens, 1829 in the family Prodoxidae Riley, 1881 for the first time, without giving arguments for the new combination. Subsequent publications followed this new taxonomic concept (e.g. Deutsch 2012; Huemer 2013; Karsholt and Nieukerken 2004–2011). However, Kurz (2002–2012), after dissection and careful examination of a female, reconsidered the taxonomy and informally recombined the species in *Incurvaria* Haworth, 1828 (Incurvariidae). This work

is now supported after an extensive analysis of characters of the male and female genitalia and cytochrome *c* oxidase subunit I (COI-5P) DNA barcodes and we therefore now formally present this revised combination.

Material and methods

Our study is based on examination of ten specimens of *I. stangei* which are all deposited in the collections of the Tiroler Landesmuseum Ferdinandeum, Natural History Collections, Hall, Austria. Genitalia preparations followed standard techniques (Robinson 1976), with some modifications, especially the implementation of the so-called unrolling technique in male genitalia (Pitkin 1986). Although genitalia of both sexes are strongly sclerotized, male preparations were slightly stained with Mercurochrome and females with Chlorazol to highlight delicate structures. For photos, genitalia of both sexes were embedded in Euparal with ventral side up, male genitalia being unrolled. Additionally, male genitalia were photographed in their natural three-dimensional shape before spreading, both from lateral and dorsal aspect. Photos of adults were taken with a Canon EOS 5D Mark III and Canon lens MP-E 65 at 2:1, using a ring flash. Genitalia photos were taken with a microscope (Wild Heerbrugg) using a 10x objective and a 2.5x ocular. All photos were edited using the software HELICON FOCUS 4.80 and ADOBE PHOTOSHOP 6.0. For sequencing a single leg was removed, placed in a 96-well lysis plate and sent for analysis to the CCDB (Canadian Centre for DNA Barcoding, University of Guelph, Canada) where DNA extraction, PCR amplification, and sequencing were performed following standard high-throughput protocols (DeWaard et al. 2008). A Neighbor-Joining (NJ) tree of 26 European species of Incurvariidae and Prodoxidae plus a species of the type genus *Prodoxus* Riley, 1880 was constructed using MEGA 7 (Kumar et al. 2016). This tree is based on 132 barcode sequences which are available in the public dataset "DS-LEPIINCU Lepidoptera - Incurvarioidea" in the Barcode of Life Datasystems BOLD (Ratnasingham and Hebert 2007). Node confidences were assessed using 500 bootstrap replicates.

Abbreviations:

NHMW Naturhistorisches Museum, Wien, Austria.

TLMF Tiroler Landesmuseum Ferdinandeum, Innsbruck, Austria.

Results

Incurvaria stangei Rebel, 1903, revised combination

Lampronia stangei (Rebel, 1903) Wojtusiak 1996: page 29.

Type material. Holotype, female: Italy, South Tyrol, Tschierspitze, Felsgebiet nördlich des Grödner Jochs, 2500 m, 31.vii.1902, leg G. Stange, NHMW [not examined, see remarks].

Material examined. Austria: 1 male, East Tyrol, Lienzer Dolomiten, Karlsbaderhütte, 2300–2400 m, 02.viii.2013, leg. H. Deutsch, TLMF, barcode sample ID BC TLMF Lep 24036; 1 male, East Tyrol, Laserzgebirge, Lavanter Almtal, 1200 m, 27.vii.2012, leg. H. Deutsch, TLMF. Slovenia: 3 male, 1 female, Goriška, Julische Alpen, Triglav, Planika-Hütte, 2400 m, 05.viii.1991, leg. H. Habeler, TLMF; 1 male, Goriška, Julis-

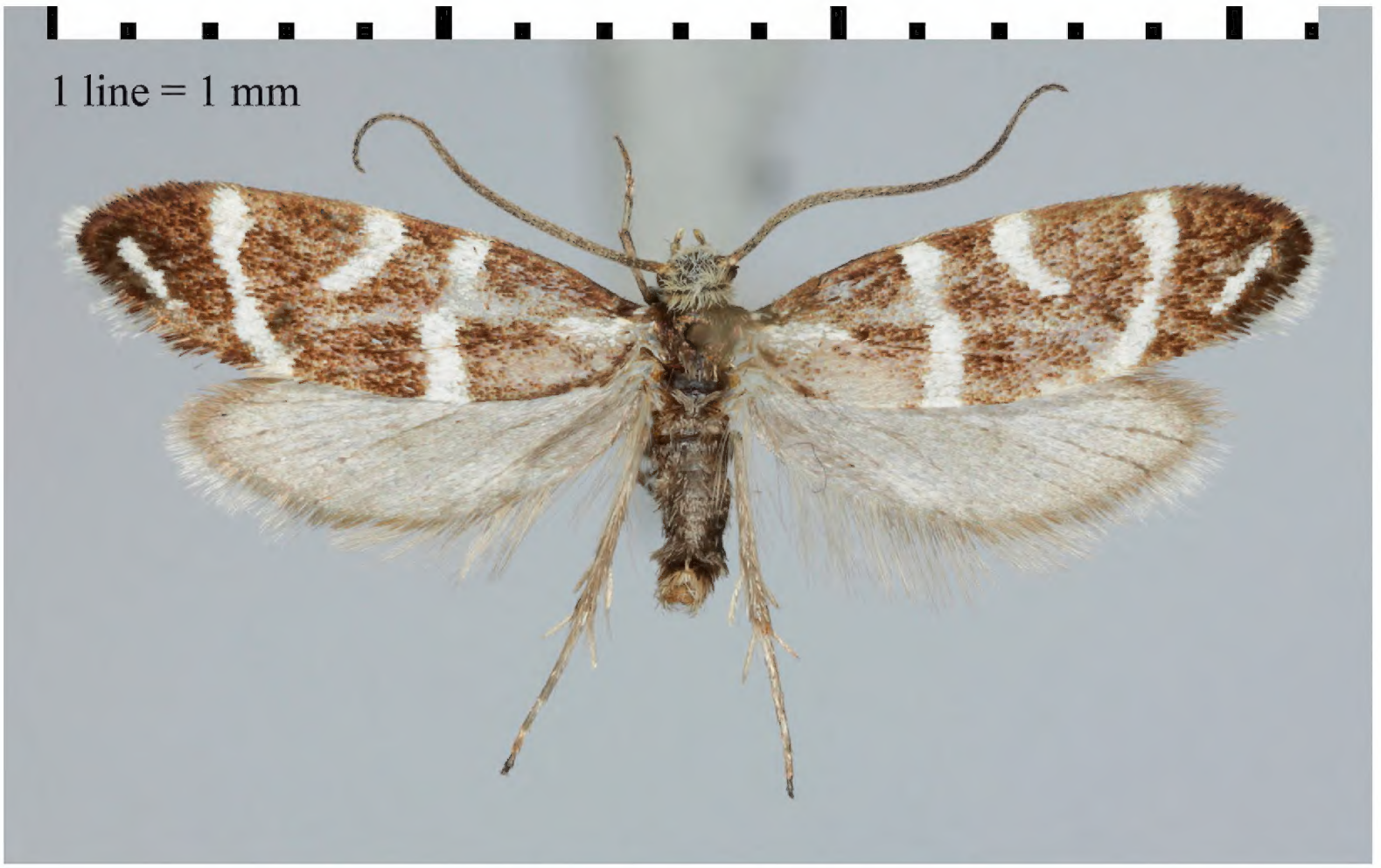

Figure 1. *Incurvaria stangei* Rebel, 1903, male, Austria, East Tyrol, Laserzgebirge, Lavanter Almtal, 1200 m, 27.vii.2012, leg. H. Deutsch.

che Alpen, Kamin, Podi, 2200–2300 m, 19.vii.1996, leg. H. Deutsch, TLMF. Italy: 1 male, Friuli-Venezia Giulia, Udine, Monte Sernio Massiv, Forcella Nuviernulis, 1700 m, 16.vii.1988, leg. P. Huemer, TLMF; 1 female, Lombardia, Bergamo, Alpi Orobie, Pizzo Arera, 2500 m, 20.vii.1992, leg. P. Huemer & G. Tarmann, TLMF; 1 male, Friuli-Venezia Giulia, Pordenone, Val Montanaia, linker Talhang/ Cimolais N, 2100 m, 23.vi.2017, leg. B. Wiesmair & P. Schattanek, TLMF, barcode sample ID BC TLMF Lep 24036.

Description. Adult (Figs 1, 2). Forewing length: male (n=8), 7.0–8.0 mm; female (n=2), 5.5–6.5 mm.

Head covered with yellowish scales, frons whitish; eyes hemispheric, black; labial palpi bright yellowish brown, apically whitish; antennae in both sexes delicately ciliated, brown, covered with small white scales in fresh specimens; thorax brownish black with golden shine; abdomen grey-brown; ground colour of forewing upper side black-brown with a distinct white wing pattern consisting of: a short longitudinal basal streak, two fasciae developed as small narrow bands, inner fascia at 1/3 of the wing length runs from costal to dorsal margin (costal nearer to wing-base), outer fascia at 3/4 is arranged in the opposite direction, costal margin at 1/2 with a subtriangular spot, a short horizontal streak in the marginal area near the apex of the wing, fringes basal brownish black, distal white. Underside with the same pattern, but more blurred. Hindwings shiny grey, fringes basally dark grey, distal part brighter grey to white; legs dirty white to beige, tibiae and tarsi brownish beige, fore-tibia without epiphysis, hind-tibia with a pair of pre-apical spurs, with a long tuft of hair-like scales and a pair of long apical spurs.

DNA barcode. BOLD (BIN): BOLD:ADI9579, (Process ID):
LEAST678-17
LEAST151-17

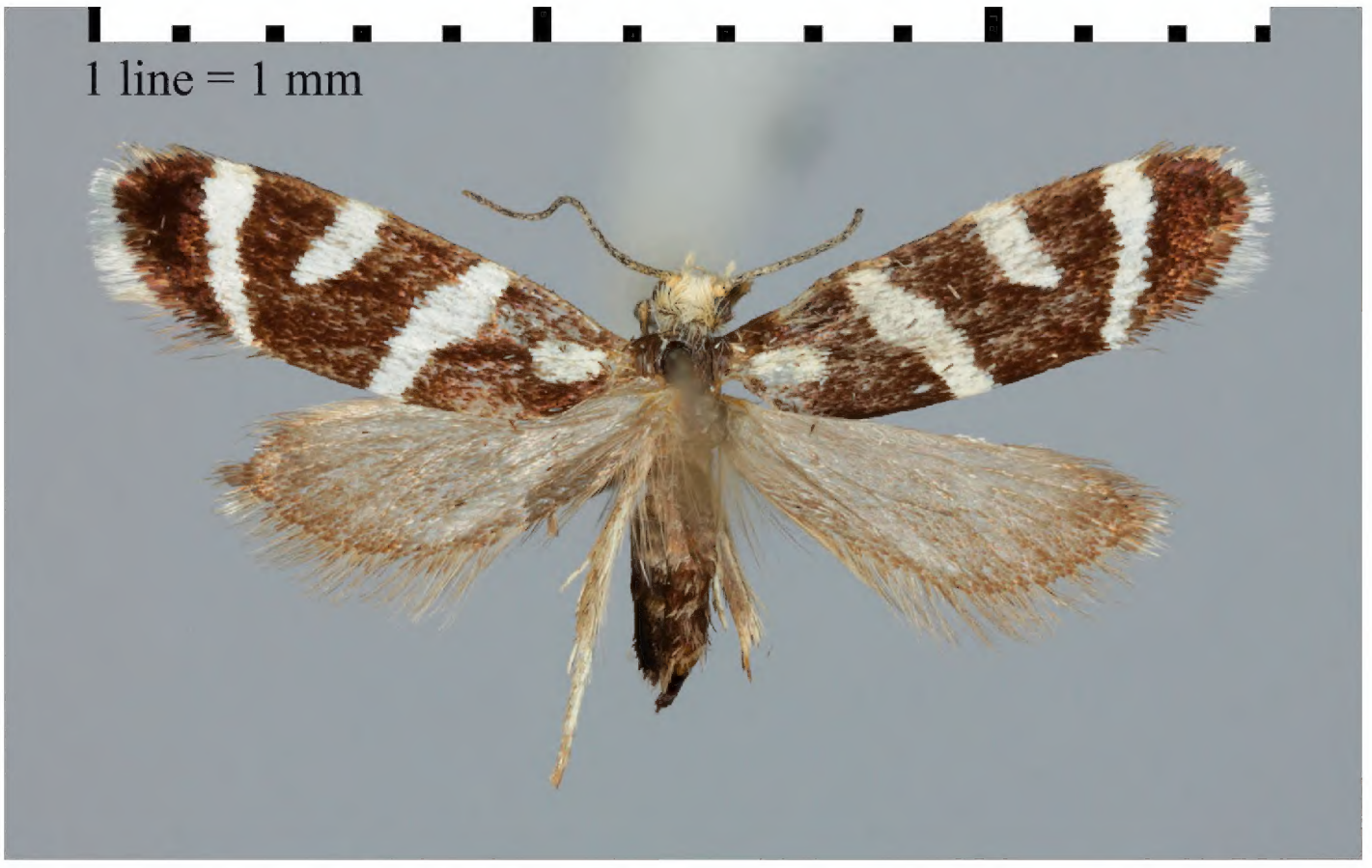

Figure 2. *Incurvaria stangei* Rebel, 1903, female, Slovenia, Goriška, Julische Alpen, Triglav, Planika-Hütte, 2400 m, 05.viii.1991, leg. H. Habeler.

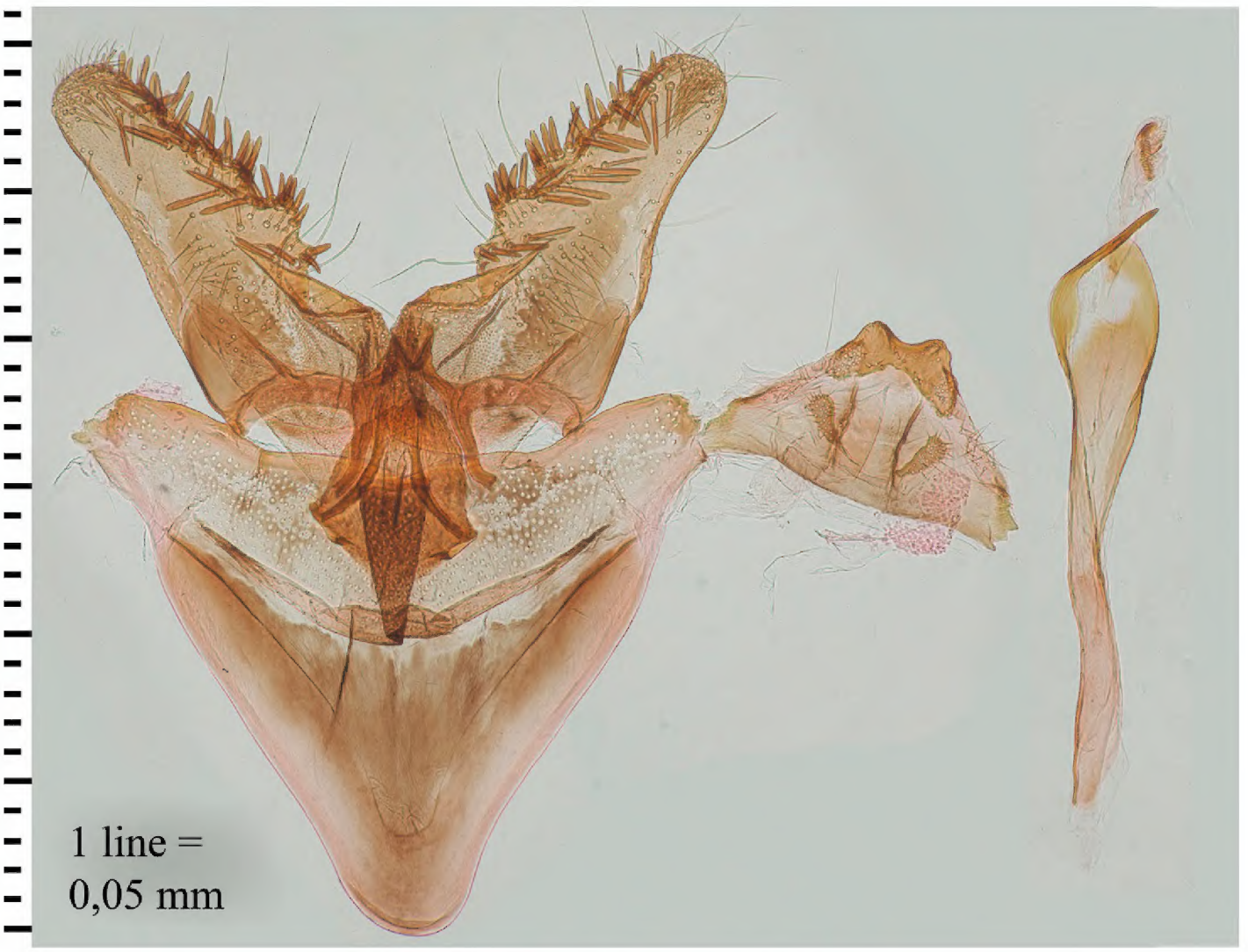

Figure 3. Unrolled male genitalia of *Incurvaria stangei* Rebel, 1903, Slovenia, Goriška, Julische Alpen, Triglav, Planika-Hütte, 2400 m, 05.viii.1991, leg. H. Habeler, Gen.Präp. m3331 P. Buchner.

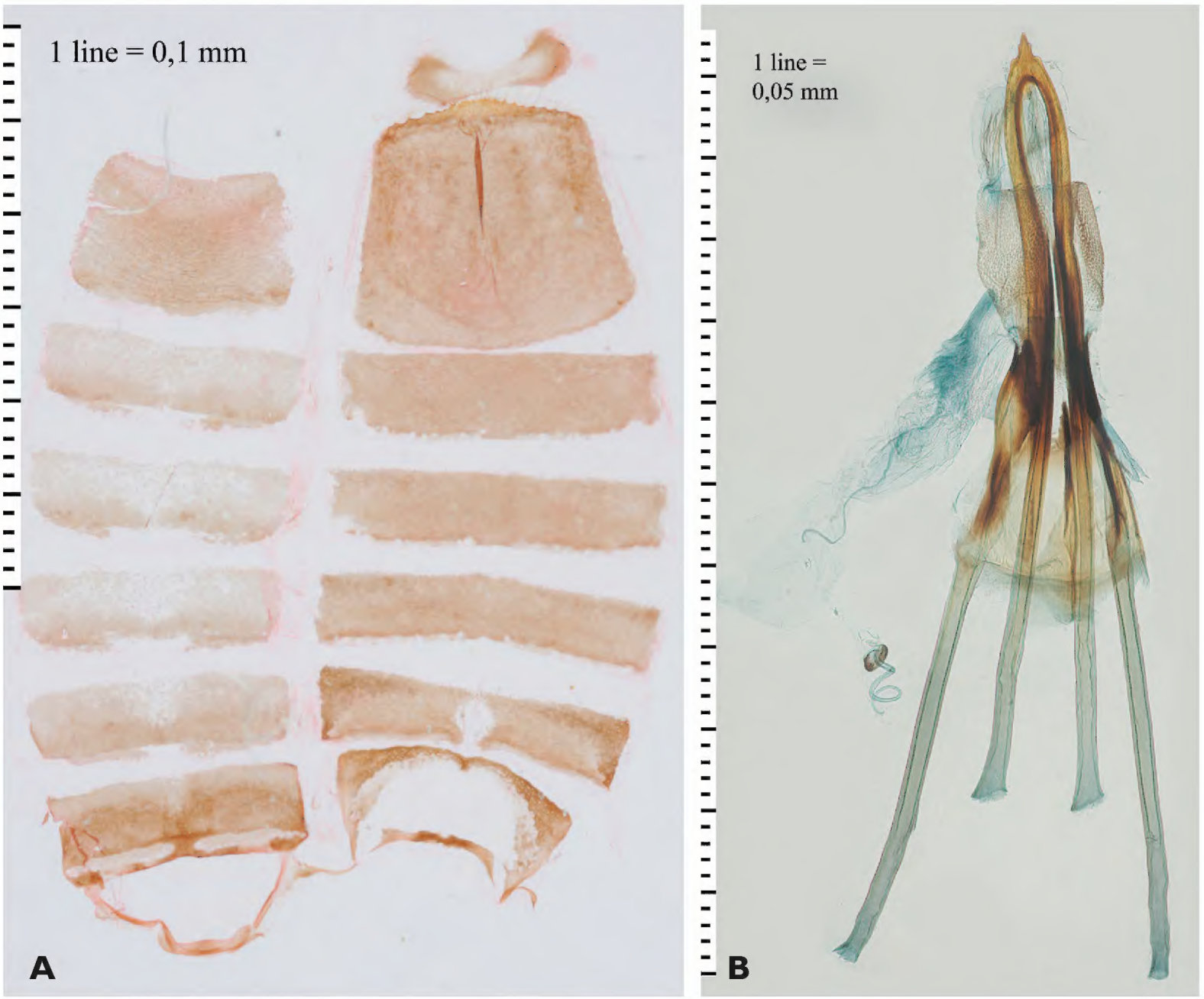

Figure 4. left: Female abdominal segments, right: Female genitalia of *Incurvaria stangei* Rebel, 1903, Slovenia, Goriška, Julische Alpen, Triglav, Planika-Hütte, 2400 m, 05.viii.1991, leg. H. Habeler, Gen.Präp. w3349 P. Buchner.

Male genitalia (Fig. 3). Uncus and tegumen short, forming a trapezoid plate, posterior margin of uncus medially concave forming a double lobed structure; socius distinctly sclerotized, club-shaped, long, distally widened; transtilla with distinct medial knob and strongly sclerotized lateral arms; sub-medial anterior projections relatively long, finger-shaped and of equal width, ending in an edge; valva short and robust; costal margin slightly concave; cucullus hirsute, with a set of distinct spines along the margin, apex hirsute and rounded; vinculum V-shaped, broad and short, distal end broad and rounded.

Female genitalia (Fig. 4). Pre-genital segments. Sternum VII as well as tergum VII of rectangular shape. Posterior part of ovipositor triangular, tip rounded, lateral margins concave with two steps; apophyses posteriores and anteriores both relatively long, with blunt ends; ductus bursae as well as bursa copulatrix short, the latter bulky and without signa; ductus spermathecae with several convolutions.

Diagnosis. Due to the unique colouration and pattern of the forewings of both sexes, *I. stangei* can be easily separated from all other members of Adeloidea.

Bionomics. The biology of *I. stangei* is unknown; the host plant as well as primary stages are still undiscovered. Huemer (1991) assumed that adults are active in the early morning hours before

Figure 5. Habitat of *Incurvaria stangei* Rebel, 1903 in Friuli-Venezia Giulia, Pordenone.

sunrise, similar to some other alpine incurvariids such as e.g. *I. vetulella* (Zetterstedt, 1839). However, Deutsch (2012) also reported attraction to artificial light. The poorly known life habits likely explain the small number of observed individuals. The adults were found in the second half of July, mainly at the border zone between the subalpine shrubs and alpine meadows from about 1700 to 2500 m elevation (Huemer 1991) (Fig. 5). Deutsch (2012), however, found a single male at a lower elevation (1250 m), which is suspected to have been drifted from its natural habitat.

Distribution. The species is endemic to the south-eastern Alps: Julian Alps: Slovenia, Goriška (Carnelutti 1978; Habeler and Gomboc 2005; Govedič and Lesar 2010; Rebel 1907); Carnic Alps: Italy, Friuli-Venezia Giulia (Huemer 1991; unpublished record), Austria, East Tyrol (Deutsch 2012); Dolomites: Italy, South Tyrol (Rebel 1903); Orobian Alps: Italy, Lombardia (unpublished record).

Remarks. Despite of a personal search by PH, the holotype could not be traced at NHMW. The Rebel collection only contains a label of the species but without any evidence of the specimen, even a pinhole is absent.

Discussion

The family concept of Incurvariidae and Prodoxidae and consequently the correct combination of genera and species remained obscure through most of the 20th century. Even in some recent literature the European genera of Prodoxidae were lumped in the family Incurvariidae (Karsholt et al.

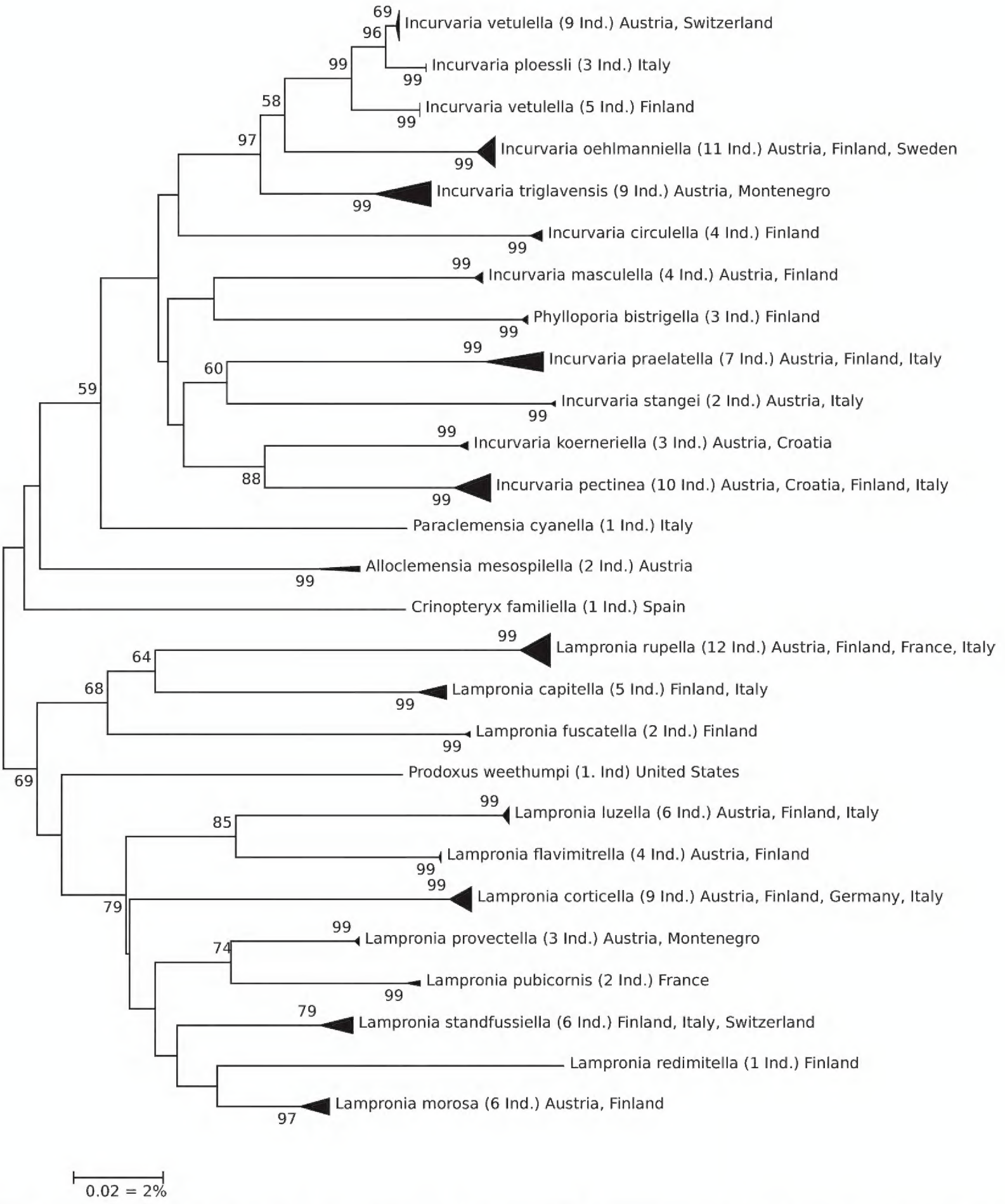
Incurvaria vetulella (9 Ind.) Austria, Switzerland
Incurvaria ploessli (3 Ind.) Italy
Incurvaria vetulella (5 Ind.) Finland
Incurvaria oehlmanniella (11 Ind.) Austria, Finland, Sweden
Incurvaria triglavensis (9 Ind.) Austria, Montenegro
Incurvaria circulella (4 Ind.) Finland
Incurvaria masculella (4 Ind.) Austria, Finland
Phylloporia bistrigella (3 Ind.) Finland
Incurvaria praelatella (7 Ind.) Austria, Finland, Italy
Incurvaria stangei (2 Ind.) Austria, Italy
Incurvaria koerneriella (3 Ind.) Austria, Croatia
Incurvaria pectinea (10 Ind.) Austria, Croatia, Finland, Italy
Paraclemensia cyanella (1 Ind.) Italy
Alloclemensia mesospilella (2 Ind.) Austria
Crinopteryx familiella (1 Ind.) Spain
Lampronia rupella (12 Ind.) Austria, Finland, France, Italy
Lampronia capitella (5 Ind.) Finland, Italy
Lampronia fuscatella (2 Ind.) Finland
Prodoxus weethumpi (1. Ind) United States
Lampronia luzella (6 Ind.) Austria, Finland, Italy
Lampronia flavimitrella (4 Ind.) Austria, Finland
Lampronia corticella (9 Ind.) Austria, Finland, Germany, Italy
Lampronia provectella (3 Ind.) Austria, Montenegro
Lampronia pubicornis (2 Ind.) France
Lampronia standfussiella (6 Ind.) Finland, Italy, Switzerland
Lampronia redimitella (1 Ind.) Finland
Lampronia morosa (6 Ind.) Austria, Finland
0.02 = 2%

Figure 6. Neighbor-Joining tree based on DNA barcodes (658 bp fragment of the mt COI gene) of European Incurvariidae and Prodoxidae. The node support values in percentages are based on 500 bootstrap replicates.

1995). However, already Nielsen (1982, 1985) convincingly separated these families based on diagnostic characters, primarily of male and female genitalia morphology. Accordingly, males of Prodoxidae can be recognized by the presence of well-defined pectinifers of the valva, whereas in the female genitalia, Prodoxidae possess a pair of stellate signa in the corpus bursae and a posteri-

orly well-rounded sternum VII and a triangular tergum VII. Furthermore, the larvae of Prodoxidae – as far as known – are endophagous in herbs and shrubs, whereas nearly all larvae of Incurvariidae are free living in self-constructed cases.

The male genitalia of *I. stangei* lack the family characteristic pectinifers typical of Prodoxidae and the more flattened, scale-shaped spines of the valva are grouped together. These characteristics support the placement of *I. stangei* in Incurvariidae. Similarly the female genitalia of *I. stangei*, which completely lack stellate signa, perfectly fit into the concept of Incurvariidae as proposed by Nielsen (1982). Furthermore, sternum and tergum VII are both of rectangular shape, characteristic for Incurvariidae. In contrast, a posteriorly well-rounded sternum VII and a triangular tergum VII is considered diagnostic for Prodoxidae (Nielsen 1982).

A Neighbor Joining tree analysis was derived from DNA barcode sequences (COI) of selected European species of Incurvariidae and Prodoxidae with *I. stangei* nesting in Incurvariidae. Considering a mean interspecific distance of 8.5% (min. 1.2%, max. 13.7%) in Incurvariidae, *I. stangei* shows a remarkable distance (12%) to its closest species, *Paraclemensia acerifoliella* (Fitch, 1854), from North America, while it is over 12.3% to *Incurvaria praelatella* (Fig. 6).

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